

**PATENT**

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

In re Application of:  
Toni A. Armstrong  
David L. De Boer

Serial No.: 10/692,762

Filed: October 24, 2003

For: METHOD FOR THE REGENERATION  
OF COTTON

Group Art Unit: 1661

Examiner: June Hwu

Atty. Dkt. No.: MONS:127USC1

**DECLARATION OF TONI A. ARMSTRONG UNDER 37 C.F.R. § 1.132**

I, Toni A. Armstrong hereby declare as follows:

1. I am a U.S. citizen and currently reside at St. Louis, MO.
2. I have been employed by Monsanto Company since July 1976, currently with the title of Research Scientist.
3. I hold a B.S. in Chemistry from the University of California. I have been conducting research in the area of agricultural biotechnology since 1976. My duties have included improving methods of plant cell culture.
4. I am a co-inventor of the above-captioned patent application, the ownership of which is assigned to Monsanto Technology LLC, and am familiar with the contents of the patent application.
5. I understand that the Patent and Trademark Office Examiner in charge of assessing the patentability of the referenced patent application has rejected the claims of the patent

application as being obvious over *Finer* (Canadian Patent Application 1,309,367; "*Finer*"), further in view of *Rangan et al.* (U.S. Patent 5,834,292; "*Rangan*").

6. The work described by *Finer* in view of *Rangan* does not teach or suggest the subject matter claimed in the present application.
7. In particular, *Finer* does not teach or suggest that use of dark lighting conditions is advantageous during a step of induction of embryogenesis of non-embryogenic cotton callus as presently claimed. This is because *Finer* states that 16:8 hr alternating light:dark conditions are preferred, and callus derived from hypocotyl tissues is unorganized, *i.e.* non-embryogenic, while callus from somatic embryos is preferred as a source of embryogenic callus. Further, the working examples of *Finer* were performed using 16:8 hr alternating light:dark conditions. *Finer* also does not state that hypocotyl-derived callus was embryogenic, nor does *Finer* demonstrate that any hypocotyl-derived callus became embryogenic. Thus *Finer* does not lead a skilled worker to the present invention.
8. Likewise, *Rangan* only used dark conditions for seed germination, and subsequently repeatedly states that further culture steps, including induction of embryogenesis, are to be performed with 16:8 hr alternating light:dark conditions (*e.g.* *Rangan* at column 7, lines 10-11; or column 8, lines 28-29). Thus *Rangan* also fails to show that dark conditions were or would be useful in improving embryogenesis. *Rangan* further serves as an example of the state of the art, with respect to lighting conditions, shortly before the filing date of the present application. Thus the combination of *Finer* and *Rangan* does not lead a skilled worker to the presently claimed invention.

9. Rangan, at column 8, lines 28-57, states that 3-5 medium changes, each lasting 3-4 weeks (*i.e.* 9-20 weeks) are required for their (not yet embryogenic) callus to form, and to cease their secretion of phenolic compounds, before the callus is then transferred for a further set of 5-7 passages, each of 3-4 weeks of growth on a second maintenance medium (*i.e.* apparently requiring up to about 15-28 additional weeks of culture), until anthocyanin pigmentation becomes evident. It is only then that development of embryogenic callus is stated to occur (*e.g.* Rangan, column 8, lines 61-62) apparently at about 24-48 weeks of culture. Similarly, Examples 1 and 3 of Rangan states that calli (not yet embryogenic) took 3-4 weeks to initially arise from cotyledon explants, and were then subcultured repeatedly while taking four to six months to form somatic embryos. Thus, when utilizing the alternating light:dark culture conditions described in Rangan, a period of cotton tissue culture spanning a minimum of more than 4 3/4- 6 months, and often longer, is apparently needed for formation of somatic embryos (*i.e.* to observe embryogenesis).
10. In contrast, the present methods claimed in this application allow for unexpected and surprising improvement in induction of embryogenesis of non-embryogenic cotton callus tissue. As summarized for instance in Example 2, the improvement relates to both the rate of embryogenesis (*e.g.* a 2- 5 fold improvement in embryogenesis frequency as compared with results seen for culture without a period of growth under dark conditions), as well as the rapidity with which embryogenesis is seen (*e.g.* within 8-10 weeks following transfer to dark conditions as described in Example 2 at the last sentence on page 25 of the Specification, and within 12 weeks of initial transfer to selective media as described at the last paragraph of Example 1).

11. None of the other references are asserted to, and in any event do not cure, this defect in Finer, or in Finer in view of Rangan, with respect to use of dark conditions to obtain embryogenic cotton callus from non-embryogenic callus tissues, as presently claimed.
12. I hereby declare that all statements made of my own knowledge are true and all statements made on information are believed to be true and further that the statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under § 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of this application or any patent issued thereon.

Date: 1/27/2010

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